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1/77

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Request for grant of a patent

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1.	Your reference	**G-32577P1/BCK 9926**
2.	Patent applicat... 0217149.4	24 JUL 2002
3	...ddress and postcode of the or of each applicant *(underline all surnames)*	**BIOCHEMIE GESELLSCHAFT MBH** **A-6250 KUNDL TIROL** **AUSTRIA**
	Patent ADP number *(if you know it)* If the applicant is a corporate body, give the country/state of its incorporation	**AUSTRIA** 83515801
4.	Title of invention	**Organic compounds**
5.	Name of your agent *(If you have one)* "Address for service" in the United Kingdom to which all correspondence should be sent *(including the postcode)*	**B.A. YORKE & CO.** **CHARTERED PATENT AGENTS** **COOMB HOUSE, 7 ST. JOHN'S ROAD** **ISLEWORTH** **MIDDLESEX TW7 6NH**
	Patents ADP number *(if you know it)*	**1800001**

6.	If you are declaring priority from one ore more earlier patent applications, give the country and the date of filing of the or of each of these earlier applications and *(if you know it)* the or each application number	Country	Priority application number *(if you know it)*	Date of filing (day/month/year)

7.	If this application is divided or otherwise derived from an earlier UK application, give the number and the filing date of the earlier application	Number of earlier application	Date of filing (day/month/year)

8.	Is a statement of inventorship and of right to grant of a patent required in support of this request? *(Answer 'Yes' if:* *a) any applicant named in part 3 is not an inventor, or* *b) there is an inventor who is not named as an applicant, or* *c) any named applicant is a corporate body.* *(see note (d))*	**Yes**

Patents Form 1/77

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Continuation sheets of this form	
Description	14
Claim(s)	2
Abstract	1
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Priority documents	
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Statement of inventorship and right to grant of a patent *(Patents Form 7/77)*	
Request for preliminary examination and search *(Patents Form 9/77)*	ONE
Request for substantive examination *(Patents Form 10/77)*	
Any other documents *(please specify)*	

11. I/We request the grant of a patent on the basis of this application

Signature B. A. Yorke & Co. Date 24 July 2002

12. Name and daytime telephone number of person to contact in the United Kingdom

Mrs. E. Cheetham
020 8560 5847

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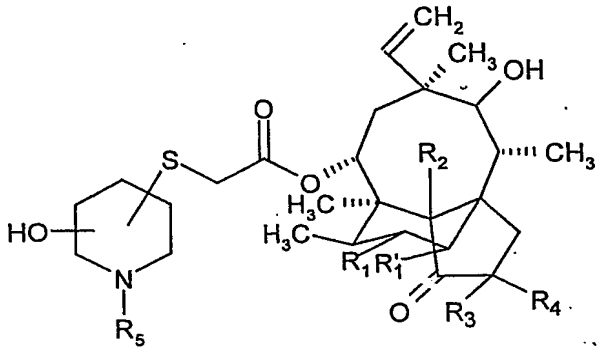
Organic Compounds

The present invention relates to organic compounds having e.g. antimicrobial, e.g. antibacterial, activity; more specifically the present invention relates to mutilins.

In one aspect the present invention provides a compound of formula



I

wherein

R_1 and R_1' are hydrogen or deuterium,

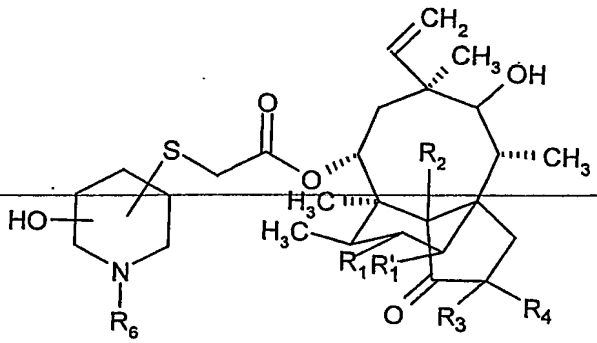
R_2, R_3 and R_4 are hydrogen or deuterium, and

R_5 is hydrogen or a residue of an amino acid.

In another aspect the present invention provides a compound of formula I selected from the group consisting of

- 14-O-[4-hydroxy-piperidin-3-yl-sulfanylacetylmutilin,
- 14-O-[3-hydroxy-piperidin-4-yl-sulfanylacetylmutilin,
- 14-O-[4-hydroxy-N-valyl-piperidin-3-yl]-sulfanylacetylmutilin, such as 14-O-[4-hydroxy-N-(R)-valyl-piperidin-3-yl]-sulfanylacetylmutilin, e.g. in the form of a hydrochloride,
- 14-O-[3-hydroxy-N-valyl-piperidin-4-yl]-sulfanylacetylmutilin, such as 14-O-[3-hydroxy-N-(R)-valyl-piperidin-4-yl]-sulfanylacetylmutilin, e.g. in the form of a hydrochloride, and
- 14-O-[3-hydroxy-N-histidinyl-piperidin-4-yl]-sulfanylacetylmutilin, such as 14-O-[3-hydroxy-N-(R)-histidinyl-piperidin-4-yl]-sulfanylacetylmutilin, e.g. in the form of a dihydrochloride.

In another aspect the present invention provides a compound of formula



II

wherein

R_1 and R_1' are hydrogen or deuterium,

R_2, R_3 and R_4 are hydrogen or deuterium, and

R_6 is a protective group, or the residue of a protected amino acid.

Protective group include protecting groups which may be, e.g. selectively, removed, if desired, and include protecting groups which are conventional in chemistry, e.g. (pleuro)mutilin chemistry, preferably tert.butoxycarbonyl (BOC), e.g. which BOC can be removed e.g. by treatment with etheric HCl.

In another aspect the present invention provides a compound of formula II selected from the group consisting of

- 14-O-[N-BOC-4-hydroxy-piperidin-3-yl-sulfanylacetylmutilin,
- 14-O-[N-BOC-3-hydroxy-piperidin-4-yl-sulfanylacetylmutilin,
- 14-O-[4-hydroxy-N-(N-BOC-valyl)-piperidin-3-yl]-sulfanylacetylmutilin, such as 14-O-[4-hydroxy-N-(N-BOC-(R)-valyl)-piperidin-3-yl]-sulfanylacetylmutilin, e.g. in the form of a hydrochloride,
- 14-O-[3-hydroxy-N-(N-BOC-valyl)-piperidin-4-yl]-sulfanylacetylmutilin, such as 14-O-[3-hydroxy-N-(N-BOC-(R)-valyl)-piperidin-4-yl]-sulfanylacetylmutilin, e.g. in the form of a hydrochloride, and
- 14-O-[3-hydroxy-N-histidinyl-piperidin-4-yl]-sulfanylacetylmutilin, such as 14-O-[3-hydroxy-N-(R)-histidinyl-piperidin-4-yl]-sulfanylacetylmutilin, e.g. in the form of a dihydrochloride.

In a compound of formula I or of formula II a carbon atom of the piperidine ring is bound to a sulphur group. That sulphur group may be in any position in the piperidine ring, e.g. in position 2, 3, 4 5 or 6, preferably in position 3 or 4. In a compound of formula I or of formula II a hydroxy group is bound to the piperidine ring. That hydroxy group may be in any position

in the piperidine ring, e.g. in position 2, 3, 4, 5 or 6, preferably in position 3 or 4. In a preferred compound of formula I or of formula II the sulphur group is in position 3 and the hydroxy group in position 4; or the sulphur group is in position 4 and the hydroxy group is in position 3 of the piperidine ring.

"A residue of an amino acid" as used herein means that in a compound of formula I the carbonyl group of said amino acid is bound to the N of the piperidine and the –OH group is missing, i.e. the N of the piperidine ring is acylated by the carboxylic group of an amino acid. Preferably the residue of an amino acid is valyl or histidinyl.

Compounds provided by the present invention, e.g. a compound of formula I or of formula II, are hereinafter designated as "compound(s) of (or compound(s) according to) the present invention". A compound of the present invention includes a compound in any form, e.g. in free form, in the form of a salt, in the form of a solvate and in the form of a salt and a solvate.

In another aspect the present invention provides a compound of formula I or of formula II in the form of a salt, or in the form of a salt and in the form of a solvate, or in the form of a solvate.

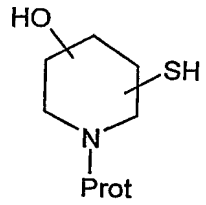
A salt of a compound of the present invention includes a pharmaceutically acceptable salt, e.g. including a metal salt or an acid addition salt. Metal salts include for example alkali or earth alkali salts; acid addition salts include salts of a compound of formula I with an acid, e.g. hydrogen fumaric acid, fumaric acid, naphthalin-1,5-sulphonic acid, hydrochloric acid, deuterochloric acid; e.g. hydrochloric acid or deuterochloric acid, preferably hydrochloric acid. A compound of the present invention may be converted into a corresponding compound in the form of a salt; and vice versa. A compound of the present invention in free form or in the form of a salt and in the form of a solvate may be converted into a corresponding compound in free form or in the form of a salt in unsolvated form; and vice versa.

A compound of of the present invention may exist in the form of isomers and mixtures thereof; e.g. optical isomers, diastereoisomers, cis trans conformers. A compound of the present invention may e.g. contain asymmetric carbon atoms and may thus exist in the form

of diastereoisomeres and mixtures thereof, e.g. racemates. For example the group bound via the sulphur atom to the piperidine ring in a compound of formula I may be in the (R)- or in the (S)-configuration or in the form of mixtures thereof. E.g. the amine group of the amino acid residue, e.g. valyl or histidinyl residue, which is acylating the nitrogen atom of the piperidene ring may be in the (S)-configuration, in the (R)-configuration or in the form of mixtures therof. Isomeric mixtures may be separated as appropriate, e.g. according to a method as conventional, to obtain pure isomers. The present invention includes a compound of the present invention in any isomeric form and in any isomeric mixture.
Preferably the cofiguration in the mutilin ring of a compound of the present invention is the same as in a naturally produced mutilin.

In another aspect the present invention provides a process for the production of a compound of formula I or of formula II comprising the steps

a) reacting a compound of formula



III

wherein Prot is a protective group, e.g. BOC, with 22-O-tosyl-pleuromutilin and tert.But-OK to obtain a compound of formula II, wherein R_6 is a protective group, e.g. BOC,

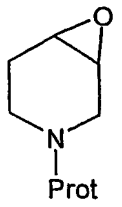
b) deprotecting the nitrogen group of the piperidinyl ring in a compound obtained in step a), e.g. by use of etheric HCl, to obtain a compound of formula I, wherein R_5 is hydrogen, optionally

c) reacting a compound obtained in step b) with an amino-protected, e.g. BOC-protected, amino acid, e.g. valine or histidine, to obtain a compound of formula II, wherein R_6 is the residue of a protected amino acid, e.g. protected valine or histidine, preferably BOC-protected valine or histidine; optionally

d) deprotecting the amino group of the amino acid residue of a compound obtained in step c) to obtain a compound of formula I, wherein R_5 is a residue of an amino acid, e.g. valyl or histidinyl; e.g. in the form of a salt, such as a hydrochloride; and optionally

e) introducing deuterium into a compound of formula I obtained in step d) to obtain a compound of formula I, wherein R_2, R_3 and R_4 are deuterium, and R_1, R'_1 and R_5 are as defined above.

In a preferred embodiment a compound of formula II, and, in consequence, e.g. according to step b) to f) of the present invention, a compound of formula I, may be obtained by reaction of a compound of formula



IV

with thiapleuromultilin and Al_2O_3 to obtain a mixture of compounds of formula II, wherein R_6 is a protective group, e.g. BOC and wherein
in one of the compounds of the mixture the hydroxy group is in position 3 and the sulphur group of the thiapleuromutilin is in position 4 of the piperidine ring, and in the other compound of the mixture the hydroxy group is in position 4 and the sulphur group of the thiapleuromutilin is in position 3 of the piperidine ring. That regioisomeric mixture may be

- separated to obtain pure compounds of formula II which pure compounds of formula II may be treated further according to steps b) to f) of the present invention to obtain pure compounds of formula I; or
- the regioisomeric mixture of compounds of formula II may be treated further according to steps b) to f) of the present invention to obtain a mixture of corresponding regioisomers of compounds of formula I which mixture may be separated to obtain pure compounds of formula I.

Separation of regioisomers may be carried out as appropriate, e.g. by chromatography.

If in step c. of to the present invention the amino acid is used in the (R)-form, e.g.(R)-valine, (R)-histidine, a compound of formula I or II is obtained, wherein the amine group of the (protected) amino acid group attached to nitrogen atom of the piperidine ring is in the (R)-configuration; and if in step c) of the present invention the amino acid is used in the (S)-form, e.g.(S)-valine, (S)-histidine, a compound of formula I or II is obtained, wherein the amine group of the (protected) amino acid group attached to nitrogen atom of the piperidine ring is in the (S)-configuration.

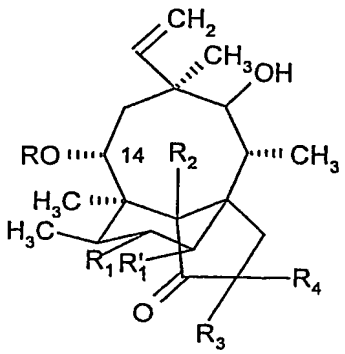
Compounds of formula II are novel and may be useful as intermediates in the production of a compound of formula I, or may be pharmaceutically active.

Protection groups include appropriate protection groups, e.g. such as useful in organic chemistry, e.g. (pleuro)mutilin chemistry, e.g. protection groups as conventional, such as BOC.

A compound of formula III is known or may be obtained according to a method as conventional. Any compound described therein may be produced according, e.g. analogously, to a process as conventional, or as described herein.

Replacement of hydrogen atoms in a compound of formula I, e.g. in the form of a salt; by deuterium atoms may be carried out as appropriate, e.g. according to a method as conventional, e.g. or according to a method described herein; e.g. by treatment of a compound of formula I with deuterochloric acid (DCl) in an appropriate solvent (system) and isolation of a compound of formula I, e.g. in the form of a salt, wherein hydrogen atoms, e.g. in the meaning of R_2, R_3 and R_4 are replaced by deuterium atoms.

The production of a compound of formula I, wherein R_1 and R'_1 is deuterium may be carried out as appropriate, e.g. according to a method as conventional, e.g. via treatment of a compound of formula



V

wherein the carbon atoms carrying R_1 and R'_1, which both are hydrogen, together form a double bond and wherein R_2, R_3 and R_4 are hydrogen, which is a known compound, with deuterium; to obtain a compound of formula V, wherein R_1 and R'_1 are deuterium and R_2, R_3 and R_4 are hydrogen; and further reacting a compound of formula V, wherein R_1 and R'_1 are deuterium and R_2, R_3 and R_4 are hydrogen as appropriate, e.g. according to a method as conventional, to obtain a compound of formula II, wherein, R_1 and R'_1 are deuterium and R_2, R_3 and R_4 are hydrogen. R may be a residue which is chemically not affected by deuterium addition, e.g. -CO-CH_2OH.

The compounds of formula I are hereinafter designated as "active compound(s) of the

present invention" which exhibit pharmacological activity and are therefore useful as pharmaceuticals. The compound of formula II may be useful intermediates, which may also exhibit pharmacological activity.

For example, the active compounds of the present invention (e.g. and compounds of formula II) show antimicrobial, e.g. antibacterial, activity against gram negative bacterias, such as Escherichia coli; and against gram positive bacteria, such as Staphylococcus aureus, Streptococcus pyogenes, Streptococcus pneumoniae, Mycoplasms, Chalmydia and obligatory anaerobes, e.g. Bacteroides fragilis; in vitro in the Agar Dilution Test or Microdilution Test according to National Commitee for Clinical Laboratory Standards (NCCLS) 1997, Document M7-A4 Vol.17, No. 2: "Methods for dilution Antimicrobial Susceptibility Tests for Bacteria that Grow Aerobically – Fourth Edition, Approved Standard" and e.g. in vivo in systemic infections in mice. The active compounds of the invention show an surprising overall activity spectrum.

In another aspect the present invention provides a compound of formula I, e.g. or of formula II, for use as a pharmaceutical, preferably as an antimicrobial, such as an antibiotic.

In a further aspect the present invention provides a compound of formula I e.g. or of formula II, for use in the preparation of a medicament for the treatment of microbial diseases, for example of diseases caused by bacteria, e.g. selected from Staphylococcus aureus, Streptococcus pyogenes, Streptococcus pneumoniae, Mycoplasms, Chalmydia e.g. and obligatory anaerobes; e.g. including penicillin or multidrug-resistant strains, e.g. of Streprococcus pneumoniae; e.g. including vancomycin-resistant strains, e.g. of Enterococcus faecium; e.g. and including methicillin-resistant strains, e.g. of Staphylococcus aureus.

In a further aspect the present invention provides a method of treatment of microbial diseases which comprises administering to a subject in need of such treatment an effective amount of a compound of formula I, e.g. or of formula II; e.g. in the form of a pharmaceutical composition.

For antimicrobial treatment, the appropriate dosage will, of course, vary depending upon, for example, the active compound of the present invention employed, the host, the mode of administration and the nature and severity of the conditions being treated. However, in

general, for satisfactory results in larger mammals, for example humans, an indicated daily dosage is in the range from about 0.5 to 3 g, of an active compound of the present invention conveniently administered, for example, in divided doses up to four times a day.
An active compound of the present invention may be administered by any conventional route, for example orally, e.g. in form of tablets or capsules, or parenterally, e.g. in the form of injectable solutions or suspensions, e.g. in analogous manner to erythromycins, such as azithromycin.

The active compounds of the present invention may be administered in the form of a pharmaceutically acceptable salt, e.g. an acid addition salt or metal salt; or in free form; optionally in the form of a solvate. The active compounds of the present invention in the form of a salt exhibit the same order of activity as the active compounds of the present invention in free form.

In another aspect the present invention provides a pharmaceutical composition comprising a compound of formula I, e.g. or of formula II, in free form or in the form of a pharmaceutically acceptable salt; e.g. and/or in the form of a solvate; in association with at least one pharmaceutical, excipient, e.g. carrier or diluent.

Such compositions may be manufactured according to a method as conventional.
Unit dosage form may contain, for example, from about 100 mg to about 1 g.

The active compounds of the present invention are additionally suitable as veterinary agents, e.g. veterinary active compounds, e.g. in the prophylaxis and in the treatment of microbial, e.g. bacterial diseases, in animals, such as fowl, pigs and calves; e.g. and for diluting fluids for artificial insemination and for egg-dipping techniques.

In another aspect the present invention provides a compound of formula I, e.g. or of formula II for use as a veterinary agent.

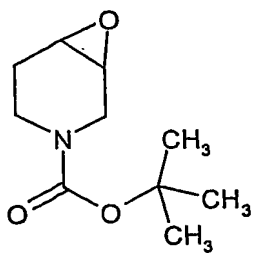
In a further aspect the present invention provides a compound of formula I, e.g. or of formula II, for the preparation of a veterinary composition which is useful as a veterinary agent.

In another aspect the present invention provides a veterinary method for the prophylaxis and in the treatment of microbial, e.g. bacterial diseases which comprises administering to a subject in need of such treatment an effective amount of a compound of formula I, e.g. or of formula II, e.g. in the form of a veterinary composition.

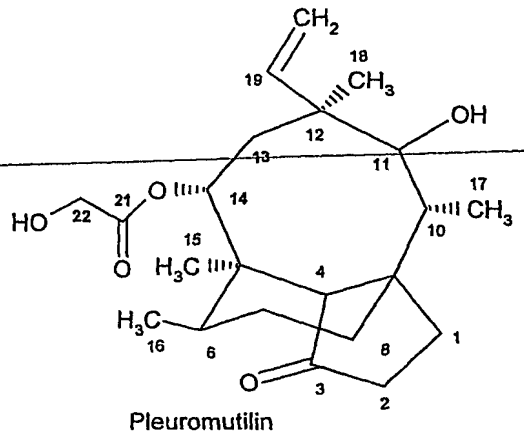
For use of the active compounds of the present invention as a veterinary agent, the dosage will of course vary depending upon the size and age of the animal and the effect desired; for example for prophylactic treatment relatively low doses would be administered over a longer time period, e.g. 1 to 3 weeks. Preferred doses in drinking water are from 0.0125 to 0.05 weight by volume, particularly 0.0125 to 0.025; and in foodstuffs from 20 to 400 g/metric ton, preferably 20 to 200 g/metric ton. It is preferred to administer the active compounds of the present invention as a veterinary agent to hens in drinking water, to pigs in foodstuff and to calves orally or parenterally, e.g. in the form of oral or paraenteral preparations.

In the following examples all references to temperature are in degrees Centigrade and are uncorrected.

The following abbreviations are used:
RT = room temperature
BOC = tert.butyloxycarbonyl
TBAF = tetrabutylammoniumfluoride
Diast. = diastereoisomer
EDC = N-(3-dimethylaminopropyl)-N-ethylcarbodiimide
EE: ethyl acetate
HOBT = hydroxybenztriazole
RT: room temperature
THF = tetrahydrofurane
tert.But-OK = tert.butoxide potassium
N-BOC-3,4-Epoxy-piperidine is a compound of formula



Pleuromutilin is a compound of formula

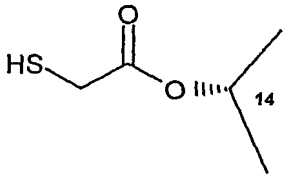


Pleuromutilin

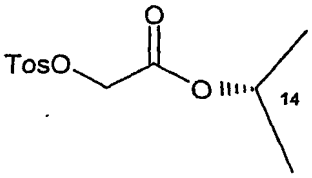
A group of formula

is a group of formula Pleuromutilin, missing the group -CO-CH_2OH.

Thiapleuromutilin is a compound of formula



22-O-Tosylpleuromutilin is a compound of formula



wherein Tos is a tosyl group.

Example 1

14-O-[N-BOC-4-Hydroxy-piperidin-3-yl]-sulfanylacetylmutilin and 14-O-[N-BOC-3-hydroxy-piperidin-4-yl]-sulfanylacetylmutilin (compounds of formula II)

40 g of (neutrally) activated Al_2O_3, moistened with THF, are treated with a solution of 1.576 g of thiapleuromutiline in 5 ml of THF and to the mixture obtained 0.398 g of N-BOC-3,4-epoxy-piperidine, dissolved in 3 ml of THF, are added. From the mixture obtained $Al_2O_3$is filtered off, from the filtrate obtained solvent is evaporated off and the evaporation residue comprising a mixture of 14-O-[N-BOC-4-hydroxy-piperidin-4-yl]-sulfanylacetylmutilin and 14-O-[N-Boc-3-hydroxy-piperidin-4-yl]-sulfanylacetylmutilin is subjected to chromatography.

0.156 g of 14-O-[N-BOC-3-Hydroxy-piperidin-4-yl]-sulfanylacetylmutilin (^{1}H-NMR ($CDCl_3$): Diast.: 4.3(b,1H,H_{II}), 4.05(m,1H,H_{VI}), 3.45(m,1H,H_{IV}), 3.28(b,2H,H_{22}), 2.8-2.6(m,2H,H_{II},H_{VI}), 2.55(m,1H,H_{III}), 1.45(s,9H,$(CH_3)_3$)); and

0.05 g of 14-O-[N-BOC-4-Hydroxy-piperidin-4-yl]-sulfanylacetylmutilin (^{1}H-NMR ($CDCl_3$): Diast.: 4.28(m,1H,H_{II}), 4.15-4.0(b,1H,H_{VI}), 3.6-3.32(b,3H,H_{11}), (1.45(s,9H,$(CH_3)_3$))

are obtained.

1.022 g of 14-O-[N-BOC-3-hydroxy-piperidin-4-yl]-sulfanylacetylmutilin are also obtained by reacting 0.466 g of N-BOC-3-hydroxy-4-mercaptopiperidine in 10 ml of THF with 0.224 g of tert.But-OK in 20 ml of THF, adding to the mixture obtained of a solution of 1.064 g of 22-O-tosylpleuromutilin in 5 ml THF, dropwise addition to the mixture obtained of 1 ml of 2-butanone and stirring at RT; and subjecting to chromatographic purification.

Example 2

14-O-[4-Hydroxy-piperidine-3-yl]-sulfanylacetylmutilin (compound of formula I)

1 mmol of 14-O-[N-BOC-4-hydroxy-piperidin-3-yl]-sulfanylacetylmutilin in 5 to 8 ml of CH_2Cl_2 is treated with 1 to 2 ml of etheric HCl, the mixture obtained is stirred at RT and 14-O-[4-hydroxy-piperidine-3-yl]-sulfanylacetylmutilin in the form of a hydrochloride precipitates and is isolated by filtration. (^{1}H-NMR ($CDCl_3$): 3.55-3.15(m,6H,H_{11},H_{22},H_{II},H_{IV},H_{VI}), 2.7-2.55(m,3H,H_{II},H_{III},H_{VI}).

Example 3

14-O-[4-Hydroxy-N-(N-BOC-(R)-valyl-piperidin-3-yl-sulfanylacetylmutilin (compound of formula II)

1.5 mmol of 14-O-[4-hydroxy-piperidine-3-yl]-sulfanylacetylmutilin dissolved in 5 ml of CH_2Cl_2 are treated with 1.5mmol HOBT, 1 mmol of (R)-valin and 1.5 mmol of EDC and stirred at RT. From the mixture obtained solvent is evaporated, the evaporation residue obtained is mixed with EE and the mixture obtained is extracted with 0.1N HCl and saturated aqueous $NaHCO_3$ solution. The organic phase obtained is dried and solvent is evaporated. 14-O-[4-Hydroxy-N-(N-BOC-(R)-valyl-piperidin-3-yl-sulfanylacetylmutilin is obtained. (^{1}H-NMR ($CDCl_3$): Rotameres/Diaster.: 5.75(m,1H,NHCO), 4.75, 4.2, 3.95 (3xm,1H,H_{II}), 4.45, 4.35(2xm,1H,NHCO), 3.55(m,1H,H_{IV}), 3.35(m,1H,H_{11}), 3.3(s,2H,H_{22}), 2.55(m,1H,H_{III}), 1.45(b,12H,$(CH_3)_3$, $(CH_3)_{15}$), 0.95, 0.7(2xm,6H,$CH(CH_3)_2$).

Example 4

14-O-[4-Hydroxy-N-(R)-valyl)-piperidine-3-yl]-sulfanylacetylmutilin (compound of formula I)

1 mmol of 14-O-[4-hydroxy-N-(N-BOC-(R)-valyl-piperidin-3-yl-sulfanylacetylmutilin in 5 to 8 ml of CH_2Cl_2 is treated with 1 to 2 ml of etheric HCl, the mixture obtained is stirred at RT and 14-O-[4-Hydroxy-N-(R)-valyl)-piperidine-3-yl]-sulfanylacetylmutilin in the form of a hydrochloride precipitates and is isolated by filtration. (^{1}H-NMR ($CDCl_3$): Diast.: 8.35(b,3H,NH_3^+), 4.5(m,2H,H_{II},NHCHCO), 3.45-3.3(m,3H,H_{11},H_{22}), 2.7, 2.55(2xm,1H,H_{III}), 3.6(m,1H,H_{IV}), 1.1(m,6H,$CH(CH_3)_2$).

Analogously to a method as described in any one of examples 1 to 4, but using appropriate starting materials, the following compounds of formula I of Examples 5 to 7 are obtained:

Example 5: 14-O-[3-hydroxy-piperidin-4-yl]-sulfanylacetylmutilin (^{1}H-NMR ($CDCl_3$): Diast. 3.4(m,1H,H_{III}) 3.35-3.3(m,4H,H_{11},H_{22},H_{VI}), 2.9(m,1H,H_{II}), 2.55(m,1H, H_{IV}).

Examle 6: 14-O-[3-Hydroxy-N-(R)-valyl-piperidin-4-yl-sulfanylacetylmutilin, e.g. in the form of a hydrochloride (^{1}H-NMR (d_6-DMSO, 350 K): Diast.: 8.05(b,3H,NH_3^+), 4.25–4.1(m, 3H, H_{II}, H_{VI},NHCHCO), 3.75(m,1H,H_{III}), 3.45-3.32(m,3H,H_{11},H_{22}), 2.89(m,1H,H_{IV}), 0.98, 0.92(2xd, 6H,$CH(CH_3)_2$, J=6 Hz).

Example 7: 14-O-[3-Hydroxy-N-(R)-histidinyl-piperidin-4-yl-sulfanylacetylmutilin, e.g. in the form of a dihydrochloride (^{1}H-NMR (d_6-DMSO, 350 K): Diast.: 8.88, 7.45(2xs,2H,

aromat.$H_{Imidazol}$), 4.75(m,1H,NHCHCO, AB-System: v_A=3.43, v_B=3.38(2H,H_{22},J=15Hz), 3.48 (d,1H, H_{11},J=6Hz), AB-System: v_A=3.23, v_B=3.15(2H,NHCHCH_2, J=8.3Hz,J=15.6Hz).

Analogously to a method as described in any one of examples 1 to 4 the following compounds of formula II of Examples 8 and 9 are obtained:

Example 8: 14-O-[3-hydroxy-N-(N-BOC-(R)-valyl)-piperidin-4-yl]-sulfanylacetylmutilin, e.g. in the form of a hydrochloride (^{1}H-NMR ($CDCl_3$): Rotameres/Diast.: 6.8, 6.68(2m,1H, NHCHCO), 5.32(m,1H,OH), 4.2(m,1H,NHCHCO), 3.85(m,1H,H_{VI}), 3.5-3.3(m,3H,H_{11},H_{22}), 3.15(m,1H,H_{III}), 2.8(m,1H,H_{IV}), 1.35(s,12H,$(CH_3)_3$,$(CH_3)_{15}$), 0.8(m,9H,CH(CH_3)$_2$),$(CH_3)_{17}$).

Example 9:14-O-[3-hydroxy-N-(N-BOC-(R)-histidinyl-piperidin-4-yl]-sulfanylacetyl-mutilin (^{1}H-NMR (d_6-DMSO, 350 K): Diast.: 8.21, 8.02(2xs,2H, aromat.$H_{Imidazol}$), 7.18(d,1H,NHCHCO,J=3.1 Hz), 6.55 (b,1H,OH), 4.65(m,1H,H_{VI}), H4,15 (m, 1H,NHCHCO), 3.5-3.1(m, 5H,NHCHCH_2,H_{11},H_{22}), 2.8(m, 1H, H_{IV}), 1.55, 1.35(2xs,18H,2x $(CH_3)_3$).

Production of starting materials

Example A - Thiapleuromutilin

a) Thiapleuromutilin in the form of the isothiuronium salt

A mixture of 106.4 g of 22-O-tosylpleuromutilin, 15.2 g of thiourea and 250 ml of aceton is refluxed for ca. 1.5 hours, cooled and from the mixture obtained solvent is evaporated and the evaporation residue is dried in vacuo. Thiapleuromutilin in the form of an isothiuronium salt is obtained.

^{1}H-NMR ($CDCl_3$): 9.82, 8.42(2xb,2H,NH_2), 7.78, 7.2(2xd,4H,arom.H_{Tosyl},J=15.8Hz).

a) Thiapleuromutilin

24.4 g of thiapleuromutilin in the form of an isothiuronium salt, dissolved in 40 ml absolute EtOH, is diluted with 70 ml of water and warmed to 90°. The mixture obtained is treated with 7.6 g of sodium disulfite in 35 ml of water and to the mixture obtained 200 ml of CH_2Cl_2 are added. The mixture obtained is heated to 90° for ca. 1.5 hours and cooled. Two phases are formed and are separated, the organic phase obtained is washed, dried, solvent is evaporated and the evaporation residue is filtered through silicagel. 8.16 g of thiapleuromutilin are obtained.

^{1}H-NMR ($CDCl_3$): 6.48(dd,1H,H_{19},$J_{19,20cis}$=11Hz,$J_{19,20trans}$=16.5Hz), 5.75(d,1H,H_{14},$J_{13,14}$= 8.5Hz), 5.38(dd,1H,H_{20},$J_{20,20}$= 1.5Hz), 5.2(dd,1H,$H_{20trans}$), 3.38(dd,1H,H_{11},$J_{11,OH}$=10.4Hz, $J_{11,10}$=6.6Hz), ABX-System: v_A=3.21, v_B=3.18,v_X=1.9 (H_{22},$J_{22,sH}$=8.2Hz,J_{AB}=15.1Hz,J_{AX}=

8.2Hz), 2.35(quint.1H,H_{10},$J_{10,17}$=8.2Hz), 2.28, 2.2(2H, $H_{H2\alpha,2\beta}$,$J_{2\alpha,2\beta}$=15.5Hz,$J_{2\alpha,1\alpha}$=$J_{2\alpha,1\beta}$= 5.5Hz), 2.19(dd,1H,H_{13},$J_{13,13}$=16Hz,$J_{13,14}$=8.5Hz), 2.12(b,1H,H_4), 1.9(t,1H,SH,$J_{22,SH}$=8.2Hz), 1.79, 176(2xq,1H,$H_{8equ.}$,$J_{7,8equ}$=3.01Hz,$J_{8,8}$=14.5Hz), 1.67(m,2H,H_1,H_6), 1.57, 1.53(2xm,1H, H_{7ax}), 1.45(s,3H,$(CH_3)_{15}$), 1.39, 1.36(2xq,1H,H_{7q},$J_{7,7}$=7.23Hz), 1.33(d,1H,$H_{13'}$), 1.18(s,3H, $(CH_3)_{18}$), 1.12(dd,1H,H_{8ax},$J_{7,8ax}$=1.14Hz), 0.89(d,3H,$(CH_3)_{17}$,$J_{10,17}$=6.54Hz), 0.74(d,3H, $(CH_3)_{16}$,$J_{6,16}$=6.5Hz). ^{1}H-NMR (d_6-DMSO): 2.85(s,1H,SH).

Example B - N-BOC-3,4-Epoxy-piperidine

a) N-BOC-1,2,5,6-tetrahydropyridine

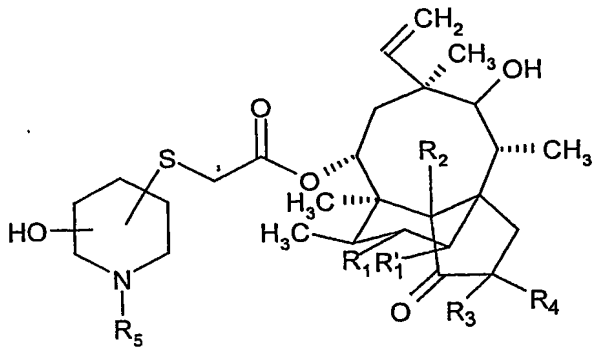
To 1.66 g of 1,2,5,6-tetrahydropyridine in 25 ml of CH_2Cl_2, 2.02 g of N-methylmorpholine are added, the mixture obtained is treated with a solution of 4.36 g $(BOC)_2O$ in 30 ml of CH_2Cl_2 and the mixture obtained is stirred for ca. 36 hours at RT. 3.4 g of N-BOC-1,2,5,6-tetrahydropyridine are obtained. ^{1}H-NMR ($CDCl_3$): 5.82(m,1H,H_{IV}), 5.64(m,1H,H_{III}), 3.86(b,2H, H_{II}), 3.47(t,2H,H_{VI}), 2.12(b,1H,H_V), 1.46(m,9H,$(CH_3)_3$).

b) N-BOC-3,4-Epoxy-piperidine

To a solution of 3.29 g of N-BOC-1,2,5,6-tetrahydropyridine in 25 ml of CH_2Cl_2, a suspension of 6.2 g of chloroperbenzoic acid in 50 ml of CH_2Cl_2 are added and the mixture obtained is stirred for ca. 12 hours at RT. The mixture obtained is extracted with saturated aqueous $NaHCO_3$-solution and 0.5 m aqueous $Na_2S_sO_3$-solution and the organic phase obtained is washed, dried and the solvent is evaporated. 3.41 g of N-BOC-3,4-epoxy-piperidine are obtained. ^{1}H-NMR ($CDCl_3$): 3.9, 3.65, 3.45, 3.1(4xm,4H,H_{II},H_{VI}), 3.28, 3.2 (2xm,2H,H_{III},H_{IV}), 2.05, 1.9(2xm,2H,H_V), 1.45(s,9H,$(CH_3)_3$).

Patent Claims:

1. A compound of formula



I

wherein

R_1 and R_1' are hydrogen or deuterium,

R_2, R_3 and R_4 are hydrogen or deuterium, and

R_5 is hydrogen or a residue of an amino acid.

2. A compound of formula I which is selected from the group consisting of

14-O-[4-hydroxy-piperidin-3-yl-sulfanylacetylmutilin,

14-O-[3-hydroxy-piperidin-4-yl-sulfanylacetylmutilin,

14-O-[4-hydroxy-N-valyl-piperidin-3-yl]-sulfanylacetylmutilin,

14-O-[4-hydroxy-N-(R)-valyl-piperidin-3-yl]-sulfanylacetylmutilin,

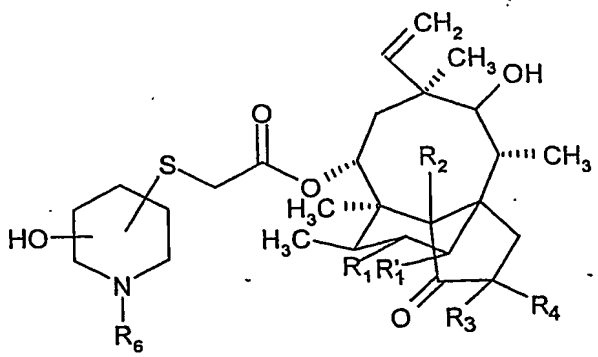
14-O-[3-hydroxy-N-valyl-piperidin-4-yl]-sulfanylacetylmutilin,

14-O-[3-hydroxy-N-(R)-valyl-piperidin-4-yl]-sulfanylacetylmutilin,

14-O-[3-hydroxy-N-histidinyl-piperidin-4-yl]-sulfanylacetylmutilin, and

14-O-[3-hydroxy-N-(R)-histidinyl-piperidin-4-yl]-sulfanylacetylmutilin.

3. A compound of formula



II

wherein

R_1 and R_1' are hydrogen or deuterium,

R_2, R_3 and R_4 are hydrogen or deuterium, and

R_6 is a protective group, or the residue of a protected amino acid.

4. A compound of formula II selected from the group consisting of
14-O-[N-BOC-4-hydroxy-piperidin-3-yl-sulfanylacetylmutilin,
14-O-[N-BOC-3-hydroxy-piperidin-4-yl-sulfanylacetylmutilin,
14-O-[4-hydroxy-N-(N-BOC-valyl)-piperidin-3-yl]-sulfanylacetylmutilin,
14-O-[4-hydroxy-N-(N-BOC-(R)-valyl)-piperidin-3-yl]-sulfanylacetylmutilin,
14-O-[3-hydroxy-N-(N-BOC-valyl)-piperidin-4-yl]-sulfanylacetylmutilin,
14-O-[3-hydroxy-N-(N-BOC-(R)-valyl)-piperidin-4-yl]-sulfanylacetylmutilin,
14-O-[3-hydroxy-N-histidinyl-piperidin-4-yl]-sulfanylacetylmutilin, and
14-O-[3-hydroxy-N-(R)-histidinyl-piperidin-4-yl]-sulfanylacetylmutilin.

5. A compound according to any one of claims 1 to 4 in the form of a salt, or in the form of a salt and in the form of a solvate, or in the form of a solvate.

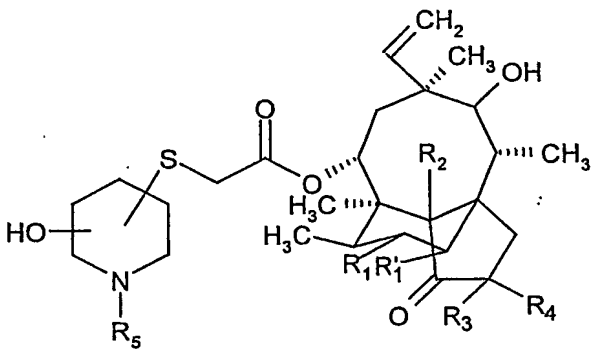
6. A compound according to any one of claims 1 to 5 for use as a pharmaceutical.

7. A method of treatment of microbial diseases comprising administering to a subject in need of such treatment an effective amount of a compound of any one of claims 1 to 5.

8. A pharmaceutical composition comprising a compound of any one of claims 1 to 5 in association with at least one pharmaceutical excipient.

Abstract

A compound of formula



I

wherein the residues have various meanings, e.g. for use as a pharmaceutical.